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SEARCH REQUEST FORM

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Scientific and Technical Inf rmation Center

Requester's Full Name: JANE ZARA Examiner #: 77512 Date: 12/19/01

Art Unit: Phone Number 30 6-5820 Serial Number: 09/241,653

Mail Box and Bldg/Room Location: 11D03 Results Format Preferred (circle): PAPER DISK E-MAIL

LEVITE12

If more than one search is submitted, please prioritize searches in order of need. MEP

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Please provide a detailed statement of the search topic, and describe as specifically as possible the subject matter to be searched. Include the elected species or structures, keywords, synonyms, acronyms, and registry numbers, and combine with the concept or utility of the invention. Define any terms that may have a special meaning. Give examples or relevant citations, authors, etc, if known. Please attach a copy of the cover sheet, pertinent claims, and abstract.

Title of Invention: Regulating hematopoiesis

Inventors (please provide full names): WAGNER et al.

Earliest Priority Filing Date: 2/2/99

For Sequence Searches Only *Please include all pertinent information (parent, child, divisional, or issued patent numbers) along with the appropriate serial number.*

Polypeptide Seq ID No: 89

Please limit to 40 Nucleotides

THANKS.

Point of Contact:
Beverly Shears
Tech. Info. Specialist
Civil Tech.
1205 Tel: 308-4994

mic 89

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STAFF USE ONLY	Type of Search	Vendors and cost where applicable
Searcher: Beverly 4994	NA Sequence (#)	STN
Searcher Phone #:	AA Sequence (#)	Dialog
Searcher Location:	Structure (#)	Questel/Orbit
Date Searcher Picked Up:	Bibliographic	Dr.Link
Date Completed: 12-28-01	Litigation	Lexis/Nexis
Searcher Prep & Review Time: 3	Fulltext	Sequence Systems
Clerical Prep Time:	Patent Family	WWW/Internet
Online Time: 20	Other	Other (specify) CGN

PTO-1590 (8-01)

ess DB(SM) 1976-2001/Dec W2
(c) 2001 The Gale Group
File 159:Cancerlit 1975-2001/Oct
(c) format only 2001 Dialog Corporation
File 164:Allied & Complementary Medicine 1984-2001/Feb
(c) 2001 BLHCIS
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File 457:The Lancet 1986-2000/Oct W1
(c) 2000 The Lancet, Ltd.
File 467:ExtraMED(tm) 2000/Dec
(c) 2001 Informania Ltd.

Set	Items	Description
S1	434	CPG(S)ANTIGEN?(S)IMMUNO?
S2	199	S1 AND MICE
S3	31	S2 AND PHOSPHOROTHIOATE
S4	12	RD (unique items)

>>>KWIC option is not available in file(s): 41, 77, 399

4/3,K/1 (Item 1 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13319312 BIOSIS NO.: 200100526461
Multiple effects of codon usage optimization on expression and immunogenicity of DNA candidate vaccines encoding the human immunodeficiency virus type 1 Gag protein.
AUTHOR: Deml Ludwig; Bojak Alexandra; Steck Stephanie; Graf Marcus; Wild Jens; Schirmbeck Reinhold; Wolf Hans; Wagner Ralf(a)
AUTHOR ADDRESS: (a)Institute for Medical Microbiology, Klinikum Regensburg, Franz-Josef-Strauss Allee 11, 93053, Regensburg: ralf.wagner@klinik.uni-regensburg.de**Germany
JOURNAL: Journal of Virology 75 (22):p10991-11001 November, 2001
MEDIUM: print
ISSN: 0022-538X
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: We have analyzed the influence of codon usage modifications on the expression levels and immunogenicity of DNA vaccines, encoding the human *immunodeficiency* virus type 1 (HIV-1) group-specific *antigen* (Gag). In the presence of Rev, an expression vector containing the wild-type (wt) gag gene flanked by essential cis-acting sites such as the ...

...in the G+C content and a Rev-independent expression and secretion of Gag in all tested mammalian cell lines, including murine C2C12 muscle cells. *Mice* immunized intramuscularly with the syngag plasmid showed Th1-driven humoral and cellular responses that were substantially higher than those obtained after injection of the Rev...

...gag and syngag vector systems with the particle gun induced a Th2-biased antibody response and no cytotoxic T lymphocytes. Deletion analysis demonstrated that the *CpG* motifs generated within syngag by codon optimization do not contribute significantly to the high immunogenicity of the syngag plasmid. Moreover, low doses of coadministered stimulatory *phosphorothioate* oligodeoxynucleotides (ODNs) had only a weak effect on antibody production, whereas at higher doses immunostimulatory and nonstimulatory ODNs showed a dose-dependent suppression of humoral responses. These results suggest that increased Gag expression, rather than modulation of *CpG*-driven vector immunity, is responsible for the enhanced immunogenicity of the syngag DNA vaccine.

4/3,K/2 (Item 1 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

09930889 Genuine Article#: 465MX No. References: 40
Title: Immune stimulation by a CpG-containing oligodeoxynucleotide is enhanced when encapsulated and delivered in lipid particles
Author(s): Mui B (REPRINT) ; Raney SG; Semple SC; Hope MJ
Corporate Source: Inex Pharmaceut Corp,100-8900 Glenlyon Pkwy,Glenlyon Business Pk/Burnaby/BC V5J 5J8/Canada/ (REPRINT); Inex Pharmaceut Corp,Burnaby/BC V5J 5J8/Canada/
Journal: JOURNAL OF PHARMACOLOGY AND EXPERIMENTAL THERAPEUTICS, 2001, V298 , N3 (SEP), P1185-1192
ISSN: 0022-3565 Publication date: 20010900
Publisher: AMER SOC PHARMACOLOGY EXPERIMENTAL THERAPEUTICS, 9650 ROCKVILLE PIKE, BETHESDA, MD 20814-3998 USA
Language: English Document Type: ARTICLE (ABSTRACT AVAILABLE)

Abstract: The therapeutic benefit from *phosphorothioate* oligodeoxynucleotides (PS ODN) containing immune stimulatory sequences (ISS) has been demonstrated in animal models of cancer and infection. In particular, when *CpG*-containing PS ODN are administered to *mice*, activation of macrophages and dendritic, NK, T, and B cells occurs, resulting in the release of an array of cytokines, including interleukin-12 (IL-12...
...152-166]. Given the propensity for SALP to target macrophages in vivo it was of interest to determine whether they could enhance the potency of *CpG* ODN to induce an immune response. In this report we show that when *CpG*-containing SALP are administered intravenously to ICR *mice* the plasma concentrations of IL-12, IFN-gamma, IL-6, monocyte chemoattractant protein-1, and TNF-alpha are greatly increased compared with the same dose...

...free ODN. The pattern of cytokine induction indicates that the immune response is T helper cell type 1-biased, similar to that observed for PS *CpG* ODN ISS in general. Furthermore, when phosphodiester (PO) ODN is substituted for PS ODN in the SALP formulation cytokine induction is even greater at the early time points, in marked contrast to free PO ODN, which is inactive. These results demonstrate that the *immunogenicity* of ISS is not only enhanced by encapsulation in lipid particles, which more closely mimic the way ISS DNA would normally be presented to *antigen* presenting cells by pathogens in vivo, but also SALP enable unmodified PO *CpG* ODN to be used as immune stimulants.
...Identifiers--BACTERIAL-DNA; IN-VITRO; *PHOSPHOROTHIOATE* OLIGODEOXYNUCLEOTIDES; C-MYC; IMMUNOSTIMULATORY OLIGODEOXYNUCLEOTIDES; ANTISENSE OLIGODEOXYNUCLEOTIDES; MODIFIED OLIGONUCLEOTIDES; CONTACT HYPERSENSITIVITY; TH1 RESPONSES; CUTTING EDGE

4/3,K/3 (Item 2 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

09882847 Genuine Article#: 460JQ No. References: 34
Title: The role of CpG sequences in the induction of anti-DNA antibodies
Author(s): Pisetsky DS (REPRINT) ; Wenk KS; Reich CF
Corporate Source: Durham VA,Durham//NC/27705 (REPRINT); Durham VA,Durham//NC/27705; Duke Univ,Med Ctr,Durham//NC/27705
Journal: CLINICAL IMMUNOLOGY, 2001, V100, N2 (AUG), P157-163
ISSN: 1521-6616 Publication date: 20010800
Publisher: ACADEMIC PRESS INC, 525 B ST, STE 1900, SAN DIEGO, CA 92101-4495 USA
Language: English Document Type: ARTICLE (ABSTRACT AVAILABLE)

Abstract: To investigate the role of *CpG* sequences in anti-DNA induction, immunization experiments were performed in *mice* to assess the *immunogenicity* of native Escherichia coli (EC) and calf thymus (CT)

in incomplete Freund's adjuvant, The effects of *CpG* sequences were further tested by comparing the adjuvant properties of a synthetic *phosphorothioate* oligonucleotide with a *CpG* motif to one with a GpC sequence. Both EC and CT DNA alone induced a limited anti-DNA response. For CT DNA, the addition of a *CpG* ODN Significantly enhanced responses whereas for EC DNA, the presence of a *CpG* oligonucleotide (ODN) or control GpC ODN did not increase responses compared to EC DNA alone. Specificity analysis by ELISA indicated that these immunizations led to the generation of cross-reactive anti-DNA autoantibodies. These results thus extend the adjuvant effects of *CpG* sequences to self *antigens* and suggest mechanisms by which self and foreign *antigens* can interact in the generation of autoimmunity. (C) 2001 Academic Press.

...Identifiers--SYSTEMIC LUPUS-ERYTHEMATOSUS; BACTERIAL-DNA; NORMAL *MICE*; SYNTHETIC OLIGONUCLEOTIDES; IMMUNOSTIMULATORY DNA; IMMUNE ACTIVATION; DSDNA ANTIBODIES; INTERFERON-GAMMA; SPECIES ORIGIN; STRANDED-DNA

4/3,K/4 (Item 3 from file: 34)

09418440 Genuine Article#: 404GZ No. References: 38

Title: Interleukin-12-and gamma interferon-dependent protection against malaria conferred by CpG oligodeoxynucleotide in *mice*

Author(s): Gramzinski RA; Doolan DL; Sedegah M; Davis HL; Krieg AM; Hoffman SL (REPRINT)
Corporate Source: USN,Med Res Ctr, Malaria Program,503 Robert Robert Grant Ave/Silver Spring//MD/20910 (REPRINT); USN,Med Res Ctr, Malaria Program,Silver Spring//MD/20910; Johns Hopkins Univ,Sch Hyg & Publ Hlth , Dept Mol Microbiol & Immunol,Baltimore//MD/21205; Univ Maryland,Sch Med, Dept Microbiol,Baltimore//MD/21201; Ottawa Civic Hosp,Loeb Res Inst,Ottawa/ON K1Y 4E9/Canada/; Univ Ottawa,Fac Hlth Sci,Ottawa/ON K1Y 4E9/Canada/; Univ Ottawa,Fac Med,Ottawa/ON K1Y 4E9/Canada/; Univ Iowa,Dept Internal Med,Iowa City//IA/52242

Journal: INFECTION AND IMMUNITY, 2001, V69, N3 (MAR), P1643-1649
ISSN: 0019-9567 Publication date: 20010300
Publisher: AMER SOC MICROBIOLOGY, 1752 N ST NW, WASHINGTON, DC 20036-2904 USA
Language: English Document Type: ARTICLE (ABSTRACT AVAILABLE)

Title: Interleukin-12-and gamma interferon-dependent protection against malaria conferred by CpG oligodeoxynucleotide in *mice*

Abstract: Unmethylated *CpG* dinucleotides in bacterial DNA or synthetic oligodeoxynucleotides (ODNs) cause B-cell proliferation and *immunoglobulin* secretion, monocyte cytokine secretion, and activation of natural killer (NK) cell lytic activity and gamma interferon (IFN-gamma) secretion in vivo and in vitro. The potent Th1-like immune activation by *CpG* ODNs suggests a possible utility for enhancing innate immunity against infectious pathogens. We therefore investigated whether the innate immune response could protect against malaria. Treatment of *mice* with *CpG* ODN 1826 (TCCATGA (CG) over bar TTCCTGA (CG) over bar, with the *CpG* dinucleotides underlined) or 1585 (ggGGTCAA (CG) over bar TTGAgggggG, with g representing diester linkages and *phosphorothioate* linkages being to the right of lowercase letters) in the absence of *antigen* 1 to 2 days prior to challenge with Plasmodium yoelii sporozoites conferred sterile protection against infection. A higher level of protection was consistently induced by *CPG* ODN 1826 compared with *CpG* ODN 1585. The protective effects of both *CpG* ODNs were dependent on interleukin-12, as well as IFN-gamma. Moreover, CD8+ T cells (but not CD4+ T cells), NK cells, and nitric oxide were implicated in the *CpG* ODN 1585-induced protection. These data establish that the protective mechanism induced by administration of *CpG* ODN 1585 in the absence of parasite *antigen* is similar in nature to the mechanism induced by immunization with radiation-attenuated P. yoelii sporozoites or with plasmid DNA encoding preerythrocytic-stage P. yoelii *antigens*. We were unable to confirm whether CD8+ T cells, NK cells, or nitric oxide

were required for the *CpG* ODN 1826-induced protection, but this may reflect differences in the potency of the ODNs rather than a real difference in the mechanism of action of the two ODNs. This is the first report that stimulation of the innate immune system by *CpG* immunostimulatory motifs can confer sterile protection against malaria.

4/3,K/5 (Item 4 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

05199397 Genuine Article#: VG398 No. References: 30
Title: AMPLIFICATION OF ANTIBODY-PRODUCTION BY *PHOSPHOROTHIOATE* OLIGODEOXYNUCLEOTIDES
Author(s): BRANDA RF; MOORE AL; LAFAYETTE AR; MATHEWS L; HONG R; ZON G; BROWN T; MCCORMACK JJ
Corporate Source: UNIV VERMONT,GENET LAB,32 N PROSPECT ST/BURLINGTON//VT/05401; UNIV VERMONT,VERMONT CANC CTR/BURLINGTON//VT/05405; LYNX THERAPEUT/HAYWARD//CA/00000; UNIV SOUTHAMPTON,DEPT CHEM/SOUTHAMPTON SO9 5NH/HANTS/ENGLAND/; UNIV VERMONT,DEPT MED/BURLINGTON//VT/05405; UNIV VERMONT,DEPT PEDIAT/BURLINGTON//VT/05405; UNIV VERMONT,DEPT PHARMACOL/BURLINGTON//VT/05405
Journal: JOURNAL OF LABORATORY AND CLINICAL MEDICINE, 1996, V128, N3 (SEP), P329-338
ISSN: 0022-2143
Language: ENGLISH Document Type: ARTICLE (Abstract Available)

Title: AMPLIFICATION OF ANTIBODY-PRODUCTION BY *PHOSPHOROTHIOATE* OLIGODEOXYNUCLEOTIDES
Abstract: A *phosphorothioate* oligodeoxynucleotide that is complementary (antisense) to the initiation region of the rev gene of HIV-1 causes hypergammaglobulinemia and splenomegaly in *mice*, and it induces B cell proliferation and differentiation in mouse spleen mononuclear cells (SMNCs) and human peripheral blood mononuclear cells in vitro. The current studies were performed to investigate the specificity of these immunomodulatory effects. Both the sense and antisense rev oligomers stimulated tritiated thymidine incorporation and secretion of *immunoglobulin* M (IgM) and *immunoglobulin* G (IgG) by mouse SMNCs in a concentration-dependent fashion, but the antisense oligomer produced greater immune effects. Studies comparing *phosphorothioate* oligomers (anti-rev, c-myc, and c-myb) either methylated or unmethylated at *CpG* dinucleotides showed that methylation effectively abrogated the proliferative effect and tended to reduce the *immunoglobulin* secretary activity, but the latter was not statistically significant except in the case of IgG in anti-rev oligomer-treated cultures. *Mice* were injected with the sense or antisense rev oligomers singly or in combination. The animals then were immunized with tetanus toroid and received a booster 21 days later. Oligodeoxynucleotide-treated *mice* had significantly higher levels of IgM antibodies on days 28 and 35 and of IgG antibodies on days 14 and 35 as compared with *mice* that were immunized but received vehicle alone. There was no evidence for additive, synergistic, or antagonistic interactions of the sense and antisense rev oligomers. These results indicate that the unmethylated anti-rev oligomer is the most potent of the *phosphorothioate* oligomers tested at activating lymphocyte proliferation and differentiation and that a single intravenous injection of this oligodeoxynucleotide augments antibody production to a specific *antigen* as long as 35 days later.

4/3,K/6 (Item 5 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

03898368 Genuine Article#: QR069 No. References: 29
Title: CPG MOTIFS IN BACTERIAL-DNA TRIGGER DIRECT B-CELL ACTIVATION
Author(s): KRIEG AM; YI AK; MATSON S; WALDSCHMIDT TJ; BISHOP GA; TEASDALE R

; KORETZKY GA; KLINMAN DM
Corporate Source: UNIV IOWA,COLL MED,DEPT INTERNAL MED/IOWA CITY//IA/52242; UNIV IOWA,COLL MED,DEPT PATHOL/IOWA CITY//IA/52242; UNIV IOWA,COLL MED,DEPT MICROBIOL/IOWA CITY//IA/52242; UNIV IOWA,COLL MED,DEPT PHYSIOL/IOWA CITY//IA/52242; VET AFFAIRS MED CTR/IOWA CITY//IA/52246; US FDA,CTR BIOL EVALUAT & RES,RETROVIRAL IMMUNOL SECT/BETHESDA//MD/20892
Journal: NATURE, 1995, V374, N6522 (APR 6), P546-549
ISSN: 0028-0836
Language: ENGLISH Document Type: ARTICLE (Abstract Available)

Abstract: UNMETHYLATED *CpG* dinucleotides are more frequent in the genomes of bacteria and viruses than of vertebrates. We report here that bacterial DNA and synthetic oligodeoxynucleotides containing unmethylated *CPG* dinucleotides induce murine B cells to proliferate and secrete *immunoglobulin* in vitro and in vivo. This activation is enhanced by simultaneous signals delivered through the *antigen* receptor. Optimal B-cell activation requires a DNA motif in which an unmethylated *CpG* dinucleotide is flanked by two 5' purines and two 3' py,rimidines. Oligodeoxynucleotides containing this *CpG* motif induce more than 95% of all spleen B cells to enter the cell cycle. These data suggest a possible evolutionary link between immune defence based on the recognition of microbial DNA and the phenomenon of *CpG* suppression' in vertebrates. The potent immune activation by *CpG* oligonucleotides has implications for the design and interpretation of studies using 'antisense' oligonucleotides and points to possible new applications as adjuvants.
...Identifiers--SYSTEMIC LUPUS-ERYTHEMATOSUS; BINDING; OLIGONUCLEOTIDES; *PHOSPHOROTHIOATE; *STIMULATION; LYMPHOCYTES; FRAGMENTS; *MICE*

4/3,K/7 (Item 1 from file: 71)
DIALOG(R)File 71:ELSEVIER BIOBASE

01571469 2000230789
Oral, intrarectal and intranasal immunizations using CpG and non-CpG oligodeoxynucleotides as adjuvants
McCluskie M.J.; Davis H.L.
ADDRESS: H.L. Davis, Loeb Health Research Institute, Ottawa Hospital, 725 Parkdale Avenue, Ottawa, Ont. K1Y 4E9, Canada
EMAIL: hdavis@lri.ca
Journal: Vaccine, 19/4-5 (413-422), 2000, United Kingdom
PUBLICATION DATE: October 15, 2000
CODEN: VACCD
ISSN: 0264-410X
PUBLISHER ITEM IDENTIFIER: S0264410X00002085
DOCUMENT TYPE: Article
LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 39

We have previously demonstrated that synthetic oligodeoxynucleotides (ODN) containing *immunostimulatory* *CpG* motifs (*CpG* ODN) are potent adjuvants in *mice* when delivered by intramuscular, intranasal and subcutaneous routes. Herein, using tetanus toxoid (TT) as a model *antigen* in BALB/c *mice*, we compared the ability of *CpG* ODN to induce mucosal and systemic humoral immune responses when *antigen* was delivered by three different routes: intrarectal, intranasal and oral. Results showed differences in immune responses with the three routes and also revealed that non-*CpG* 'control' ODN had adjuvant effects when used at mucosal sites. This was unexpected since non-*CpG* ODN do not have such *immunostimulatory* effects in vitro or after parenteral immunization. These findings were further investigated after oral delivery of a killed influenza vaccine on its own as well as combined with TT and hepatitis B surface *antigen*. Our findings demonstrate that with mucosal delivery, there is a Th2 *immunostimulatory* effect associated with the *phosphorothioate* ODN backbone, and that the presence of *CpG* motifs shifts this towards a Th1 response. (C) 2000 Elsevier Science Ltd.

4/3,K/8 (Item 2 from file: 71)
DIALOG(R)File 71:ELSEVIER BIOBASE

01469877 2000142057
Successful treatment of intracranial gliomas in rat by oligodeoxynucleotides containing CpG motifs
Carpentier A.F.; Xie J.; Mokhtari K.; Delattre J.-Y.
ADDRESS: A.F. Carpentier, Federation de Neurologie Mazarin, Hopital de la Salpetriere, 47 Boulevard de l'hopital, 75013 Paris, France
EMAIL: antoine.carpentier@psl.ap-hop-paris.fr
Journal: Clinical Cancer Research, 6/6 (2469-2473), 2000, United States
CODEN: CCREF
ISSN: 1078-0432
DOCUMENT TYPE: Article
LANGUAGES: English SUMMARY LANGUAGES: English
NO. OF REFERENCES: 25

Phosphorothioate oligodeoxynucleotides with *CpG* motifs (*CpG*-ODNs) activate various immune cell subsets and induce production of numerous cytokines. To evaluate whether *CpG*-ODNs can induce rejection of established tumors, Lewis rats were inoculated intracerebrally with syngeneic CNS-1 glioma cells and subsequently injected with *CpG*-ODNs into the tumor bed. Although all of the control rats (n = 14) died within 23 days, 88% of the animals (n = 8) treated with a single *CpG*-ODN injection 5 days after tumor inoculation showed long-term survival (>90 days; P < 0.002). *CpG*-ODNs increased tumoral infiltration with macrophage/microglial cells, CD8, and natural killer lymphocytes. *CpG* -ODN-cured animals were further protected against a second tumor challenge. *CpG*-ODNs had no effect on a s.c. CNS1 tumor in nude *mice*, which suggested that *CpG*-ODN is not directly cytotoxic and that *immunostimulation* is required for the antitumoral effect. These findings suggest that intratumoral injections of *CpG*-ODNs represent a new *immunotherapeutic* approach in human gliomas, which overcome the need for the selection and purification of a tumoral *antigen*.

4/3,K/9 (Item 1 from file: 357)
DIALOG(R)File 357:Derwent Biotechnology Abs

0268095 DBA Accession No.: 2001-07849 PATENT
Immunostimulatory composition useful for stimulating immune response in a subject, comprises antigen and immunostimulatory nucleic acid comprising oligonucleotides having unmethylated cytosine-guanine dinucleotides - oligonucleotide preparation, immunization in mouse, antisense oligonucleotide and DNA probe for nucleic acid vaccine and cancer or virus, fungus, bacterium and parasite infection gene therapy
AUTHOR: Krieg A M; Klinman D; Steinberg A D
CORPORATE SOURCE: Iowa City, IA, USA; Washington, DC, USA.
PATENT ASSIGNEE: Univ.Iowa-Res.Found.; Coley-Pharmaceutical 2001
PATENT NUMBER: US 6194388 PATENT DATE: 20010227 WPI ACCESSION NO.: 2001-217934 (2022)
PRIORITY APPLIC. NO.: US 386063 APPLIC. DATE: 19950207
NATIONAL APPLIC. NO.: US 386063 APPLIC. DATE: 19950207
LANGUAGE: English

ABSTRACT: A composition (I) containing an *immunostimulatory* DNA having a DNA sequence containing unmethylated cytosine-guanine (*CpG*) dinucleotides and an *antigen*, is claimed. To determine whether *CpG* oligonucleotides can cause in vivo immune stimulation, DBA/2 *mice* were injected i.p. with phosphate-buffered saline (PBS) or *phosphorothioate* *CpG* or non-*CpG* oligonucleotides at a dose of 33 mg/kg. Spleen cells from *mice* were examined 24 hr after oligonucleotide injection for expression of B-lymphocytes (BLs) surface activation markers Ly-6A/E, Bla-I and class II major...

...using three color flow cytometry and for their spontaneous proliferation using 3H thymidine. Expression of all three activation markers was significantly increased in BLs from *mice* injected with *CpG* oligonucleotides, but not from *mice* injected with PBS or non-*CpG* oligonucleotides. Spontaneous 3H thymidine incorporation was increased by 2- to 6-fold in spleen cells from *mice* injected with the stimulatory oligonucleotide. (I) is useful as a nucleic acid vaccine for the treatment and prevention of cancer or infections. (20pp)

4/3,K/10 (Item 2 from file: 357)
DIALOG(R)File 357:Derwent Biotechnology Abs

0264749 DBA Accession No.: 2001-04503
The potential of oligodeoxynucleotides as mucosal and parenteral adjuvants - recombinant hepatitis B virus surface antigen or tetanus toxoid, oligonucleotide adjuvant and immunization in mouse for recombinant vaccine, disease therapy and immunostimulant (conference paper)
AUTHOR: McCluskie M J; Weeratna R D; +Davis H L
CORPORATE AFFILIATE: Ottawa-Hosp.Loeb-Health-Res.Inst. Univ.Ottawa Coley-Pharmaceutical
CORPORATE SOURCE: Loeb Health Research Institute at the Ottawa Hospital, 725 Parkdale Avenue, Ottawa K1Y 4E9, Canada. email:hdavis@lri.ca
JOURNAL: Vaccine (19, 17-19, 2657-60) 2001
ISSN: 0264-410X CODEN: VACCDE
CONFERENCE PROCEEDINGS: Millenium Second World Congression Vaccines and Immunization, Liege, Belgium, 29th August-3rd September 2000
LANGUAGE: English

ABSTRACT: The potential of oligonucleotides (*CpG*, non-*CpG*, poly-T or poly-CG) as mucosal and parenteral adjuvants, was evaluated. All experiments were carried out using female BALB/c *mice*, aged 6-8 wk old, with 5-10 *mice*, with 5-10 *mice* per experimental or control group. For all immunizations, *mice* were anesthetized with halothane. Recombinant hepatitis B virus surface *antigen* (HBsAg) or tetanus toxoid were used as *antigens* for immunizations. The oligonucleotides used were synthesized with a nuclease-resistant *phosphorothioate* backbone. For oral vaccine delivery, *mice* were immunized on days 0, 1, 2 with 10 ug TT, alone or combined with 10 ug oligonucleotide. For i.m. delivery, *mice* were immunized with 1 ug HBsAg, alone or combined with 10 ug oligonucleotide, injected into the left tibialis anterior muscle. *Antigen* -specific antibodies in collected samples were detected and quantified by end-point dilution ELISA. The results showed that with mucosal delivery, there was a Th2-biased *immunostimulatory* effect that is associated with non-*CpG* oligonucleotide, and that the presence of *CpG* motifs can shift this toward a Th1 response. (17 ref)

4/3,K/11 (Item 3 from file: 357)
DIALOG(R)File 357:Derwent Biotechnology Abs

0263678 DBA Accession No.: 2001-01035
CpG DNA is an effective oral adjuvant to protein antigens in *mice* - oligonucleotide containing immunostimulatory CpG motif useful as vaccine adjuvant
AUTHOR: McCluskie M J; Weeratna R D; Krieg A M; +Davis H L
CORPORATE AFFILIATE: Loeb-Health-Res.Inst.Ottawa Univ.Iowa Univ.Ottawa Coley-Pharmaceutical
CORPORATE SOURCE: Loeb Health Research Institute at the Ottawa Hospital, 725 Parkdale Avenue, Ottawa, Ontario, Canada K1Y 4E9 email:hdavis@lri.ca
JOURNAL: Vaccine (19, 7-8, 950-57) 2000
ISSN: 0264-410X CODEN: VACCDE
LANGUAGE: English

CpG DNA is an effective oral adjuvant to protein antigens in *mice*
ABSTRACT: *CpG* DNA, an effective oral adjuvant to protein *antigens* in *mice*, was studied. Groups of female BALB/c *mice* 8-10 weeks were immunized at day 0, 7 and 14 day by oral administration of 100-ug hepatitis B virus surface *antigen* (HBsAg) or tetanus toxoid (TT), alone or combined with 50, 100, or 500 ug of oligonucleotide containing *immunostimulatory* (*CpG*) made with a nuclease-resistant *phosphorothioate*. Control group *mice* were immunized with 100-ug TT with the non-*CpG* control oligonucleotide. All samples were collected over a 2 day period 1 week after third and final immunization. The results showed that oral delivery of HBsAg without adjuvant resulted in none or only anti-HBs *immunoglobulin* (Ig) G titers in the plasma of *mice* . In contrast, much high levels of anti-HBs IgG antibodies were detected when *CpG* was added, with highest titers and lowest variability being obtained with the 100-ug dose. When TT was used as *antigen*, TT-specifc IgG titers in plasma were from 15-20-fold higher than for any of three doses of *CpG* ODN than for TT alone. Results from this study indicate that stimulatory *CpG* ODN may be effective as adjuvant with oral vaccines. (30 ref)
DESCRIPTORS: *phosphorothioate* oligonucleotide *immunostimulatory* *CpG* motif, hepatitis B virus *antigen*, tetanus toxoid, appl. vaccine adjuvant DNA sequence (Vol.20, No.3)

4/3,K/12 (Item 1 from file: 370)

00509106 (USE 9 FOR FULLTEXT)
Chlamydia Infections and Heart Disease Linked Through Antigenic Mimicry
Bachmaier, Kurt; Neu, Nikolaus; de la Maza, Luis M.; Pal, Sukumar; Hessel, Andrew; Penninger, Josef M.<CRF RID="C1">
Amgen Institute, Ontario Cancer Institute, Departments of Medical Biophysics and Immunology, University of Toronto, Toronto, Ontario M5G 2C1, Canada. Department of Pediatrics, University of Innsbruck, Medical School, Innsbruck A-6020, Austria. Department of Pathology, University of California, Irvine, CA 92697-4800, USA.
Science Vol. 283 5406 pp. 1335
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(THIS IS THE FULLTEXT)

...Abstract: the 60-kilodalton cysteine-rich outer membrane proteins of Chlamydia pneumoniae, C. psittaci, and C. trachomatis was shown to induce autoimmune inflammatory heart disease in *mice*. Injection of the homologous Chlamydia peptides into *mice* also induced perivascular inflammation, fibrotic changes, and blood vessel occlusion in the heart, as well as triggering T and B cell reactivity to the homologous...

...Text: Inflammatory heart diseases and dilated cardiomyopathy in humans can be reproduced in *mice* by immunization with heart muscle myosin (B6) . Cardiac myosin-induced autoimmune myocarditis is dependent on CD4.sup(+) T cells that recognize a heart muscle-specific...

...histocompatibilty complex (MHC) class II molecules (B7) . Various peptides of the a myosin heavy chain protein have been identified that can induce autoimmune myocarditis in *mice* (B8) (B9...

...amino acids 614 to 643) of the cardiac-specific a myosin heavy chain molecule [amhc(614-643)] induces severe inflammatory heart disease in BALB/c *mice* (B8) . The first 16 amino acids [amhc(614-629), SLKLMATLFSTYASAD] constituted a dominant autoaggressive epitope that was designated M7Aa (Table 1 and Fig. 1A) (B10...

...into M7Aa further revealed that the residues xxxMAxxxSTxxx (where x is any amino acid) were important for the pathogenicity of M7Aa in vivo (B11) . These *immunogenic* amino acids are conserved between murine and human a myosin heavy chains, and injection of the human M7Aa homolog into BALB/c *mice* also induced inflammatory heart disease (B11...
...We tested the possibility of *antigenic* mimicry between Chlamydia peptides and the M7Aa motif in our murine model of *antigen*-induced inflammatory heart disease. We immunized BALB/c *mice* with murine M7Aa or the homologous 60-kD CRP or p11-derived peptides in Freund's complete adjuvant (FCA) (B10) . All of the Chlamydia-derived peptides induced inflammatory heart disease at a similar frequency, although at significantly lower severity, as compared with M7Aa-immunized *mice* (Table 1). Like the inflammation that follows immunization with the endogenous autoantigen M7Aa, the disease induced by all the Chlamydia-derived peptides was characterized by perivascular and pericardial infiltration of mononuclear cells and fibrotic changes (Fig. 1, A, C, and D). *Immunohistochemical* characterization revealed that the inflammatory infiltrate in ChTR1 peptide-induced heart disease was similar to cardiac myosin- and cardiac myosin-derived peptide-induced myocarditis and...

...B16) (B17) . Inflammation was restricted to the heart and was not observed in skeletal muscle, lung, liver, pancreas, kidney, intestine, or uterus of peptide-immunized *mice*. Injection of *mice* with human *immunodeficiency* virus-2 [gp160 (371-383), INFIGPGKGSDPE]- or parainfluenza virus 1 [HT83b hemagglutinin-neuraminidase (291-309), DLVFDILDLKGKTKSPRYK]-derived peptides that shared homology with other *immunogenic* regions of the mouse amhc molecule [amhc (735-747), GQFIDSGKGAEKL, and amhc (314-332), DSAFDVLSFTAEEKAGVYK] did not cause inflammatory heart disease (B8) (B11) . Thus, *antigenic* mimicry between Chlamydia peptides and a heart muscle-specific myosin peptide can lead to the development of inflammatory heart disease...

...The development of murine autoimmune myocarditis depends on the activation of CD4.sup(+) T cells (B7) . To directly address the hypothesis of *antigenic* mimicry between an endogenous cardiac specific peptide and Chlamydia-derived peptides, we immunized BALB/c *mice* with M7Aa, ChTR1, or another cardiac-specific amhc-derived peptide, designated kka (B10) . This kka peptide is restricted to I-A.sup(k) MHC class II molecules, and kka immunization induces myocarditis in A/J (I-A.sup(k)) *mice* (B9) but not in BALB/c (I-A.sup(d)) *mice* (B11) . Immunization with M7Aa or ChTR1, but not with kka or FCA alone, led to splenomegaly and large expansion of TCRa (beta) .sup(+) CD4.sup...

...CD4.sup(+) and CD8.sup(+) T cells expressed CD69 and CD25, indicating that these cells had been activated in vivo (B19) . Splenic T cells from *mice* immunized with the endogenous peptide M7Aa proliferated when incubated with splenocytes pulsed with the M7Aa peptide (Fig. 2A) (B20) . Splenic T cells from these *mice* also showed a strong proliferative response to the C. trachomatis-derived peptide ChTR1 (Fig. 2A). T cells from M7Aa-or ChTR1-immunized *mice* did not proliferate above control when incubated with (gamma) -irradiated splenocytes pulsed with the nonpathogenic kka peptide. Splenic T cells from *mice* immunized with ChTR1 proliferated to ChTR1 and to the endogenous M7Aa peptide. Splenic T cells from control *mice* immunized with FCA only did not proliferate when activated with M7Aa. ChTR1, or kka. Thus, ChTR1 peptide immunizations can cross-prime for T cell reactivity...

...Cardiac myosin-induced autoimmune myocarditis can be transferred adoptively into nonimmunized recipient *mice* (B7) . To establish the autoimmune basis of Chlamydia peptide-induced heart disease, we injected splenic T cells from ChTR1-immunized *mice*, restimulated in vitro with ChTR1 peptide and murine recombinant interleukin-2 (mrIL-2), into syngeneic BALB/c recipient *mice* (four *mice* per group) (B21) . All animals developed inflammatory heart disease similar (severity 1.0 +/- 0.0) to that seen after direct immunization with ChTR1 peptide (Fig...

...stimulated in vitro with ChTR1 pepide and mrIL-2, did not induce myocarditis. Thus, ChTR1 peptide-induced myocardits can be transferred

adoptively into nonimmunized recipient *mice*.

...

...2B) (B23) . Likewise, immunization with the C. trachomatis-derived peptide ChTR1 induced the production of serum antibodies to ChTR1 and to the endogenous M7Aa peptide. *Mice* immunized with M7Aa or ChTR1 also produced antibodies to the kka peptide (Fig. 2B), suggesting that M7Aa-and ChTR1-induced heart disease leads to epitope...

...the lung or reproductive organs lead to the development of myocarditis? In our experimental model of inflammatory heart disease we used FCA as a potent *immunoactivator*. Bacterial DNA, but not mammalian DNA, has direct *immunostimulatory* effects in vitro and in vivo (B24) . We tested whether bacterial DNA--derived synthetic oligodeoxynucleotides (ODNs) containing unmethylated *CpG* islands could act as adjuvant for peptide-mediated autoimmunity. Various synthetic *CpG* motif-containing ODNs could trigger inflammatory autoimmune heart disease in M7Aa peptide-immunized BALB/c *mice* (Table 2 and Fig. 1F) (B25) . Immunization of BALB/c *mice* with a *CpG* ODN derived from the C. trachomatis CRP gene plus the M7Aa autoantigen induced inflammatory heart disease in the absence of FCA (Table 2 and Fig. 1F) (B25) . Immunizations in which a control non-*CpG* ODN was used plus peptide did not induce disease (Table 2). Thus, *CpG* motif-containing bacterial DNA, including Chlamydia DNA, can function as potent *immunoactivator* for autoimmunity...
...Chlamydia pneumoniae has been linked to atherosclerosis and the clogging of blood vessels (B3) (B26) . Experimental C. pneumoniae infections in rabbits and *mice* accelerate atherosclerosis and lead to focal periarteritis (B27) and C. trachomatis infections lead directly to myocarditis (B28) . *Mice* immunized with Chlamydia peptides developed perivascular fibrosis (Fig. 3, A and B), fibrinous occlusions of cardiac blood vessels (Fig. 3, C and D), and thickening...

...occlusion originating from blood vessel endothelium (Fig. 3C), a minimum of one per individual heart, occurred in 19 out of 32 (60%) hearts analyzed from *mice* immunized with Chlamydia-derived peptides. Similarly, fibrinous occlusion originating from blood vessel endothelium occurred in 14 out of 21 (67%) hearts analyzed from *mice* immunized with M7Aa. No fibrinous occlusions were detected in hearts from *mice* immunized with FCA only... absence of an overt bacterial infection, we then determined whether actual Chlamydia infections would lead to the activation of autoaggressive lymphocytes reactive to heart-specific *antigens*. BALB/c *mice* were infected with C. trachomatis through the respiratory tract and the reproductive organs (B31) . Inflammation of both the respiratory tract or the reproductive organs led to the production of *immunoglobulin* G (IgG) antibodies to heart-specific epitopes in BALB/c *mice* (Fig. 4). Because in the mouse model of autoimmune myocarditis, the production of IgG antibodies to heart-specific epitopes is dependent on the activation of autoaggressive T and B cells (B8) , these data show that infection by C. trachomatis can activate autoaggressive lymphocytes in BALB/c *mice*.

...

...followed by systemic activation of autoreactive T and B lymphocytes. Because Chlamydia peptides can mimic the effects of heart muscle a myosin heavy chain-derived *immunogenic* epitopes, T cells activated by Chlamydia-derived peptides may trigger organ-specific inflammation within the heart. Dendritic cells, which are resident within the heart and...

...In *mice*, the development of peptide-triggered inflammatory heart disease is related to genetic differences among inbred mouse strains (B6) . Similarly, genetic and environmental risk factors may...

...Chlamydia infections are common, and most people can expect to experience a Chlamydia infection at least once during their lifetime (B32) . Our data suggest that *antigenic* mimicry of autoaggressive myosin epitopes by peptides present not only in C. pneumoniae but also in C. trachomatis and C. psittaci may be linked to...

...of inflammatory cytokines, bystander activation of lymphocytes, or both (B34) . Our results provide experimental in vivo and in vitro evidence of

molecular mimicry between bacterial *antigens* and heart-specific proteins and indicate that bacterial peptides can trigger tissue-specific inflammation of the heart. In particular, this study establishes a causal link...

...the nonimmunogenic mouse M7A (beta) motif. Prevalence and severity of inflammatory heart disease as determined with these peptides are indicated. Six-week-old BALB/c *mice* were immunized twice at a 7-day interval with the indicated peptides (50 (mu) g per mouse] in FCA and analyzed 21 days after the...

...the nonimmunogenic mouse M7A (beta) motif. Prevalence and severity of inflammatory heart disease as determined with these peptides are indicated. Six-week-old BALB/c *mice* were immunized twice at a 7-day interval with the indicated peptides (50 (mu) g per mouse] in FCA and analyzed 21 days after the...

...severity +/- SD are indicated Reference B6 Reference B10 .

End Table: Columns 6 - 6 of 6

Figure F1

Caption: Inflammatory heart disease in BALB/c *mice* that were immunized with (A) the endogenous mouse M7Aa peptide from the a myosin heavy chain, (B) the control endogenous M7A (beta) peptide from the...

...D) the 60-kD CRP-derived peptide from C. pneumoniae (ChPN) (B10) . (E) Adoptive transfer of ChTR1 peptide-induced inflammatory heart disease into nonimmunized recipient *mice* (B21) . (F) Induction of inflammatory autoimmune heart disease in BALB/c *mice* with C. trachomatis DNA-derived CpG containing ODN as adjuvant (B25) . Perivascular inflammation is apparent in (A), (C), (D), and (F). (B) shows normal heart...

...Caption: (A) Splenic T cell proliferation and (B) serum IgG antibody production. (A) Proliferative responses to M7Aa, ChTR1, or kka peptides. Splenic T cells from *mice* immunized with the indicated peptides were cultured with (gamma) -irradiated syngeneic splenocytes pulsed with the indicated peptides (B20) . [.sup(3)H]thymidine uptake (counts per...

...IgG antibodies reactive to cardiac-specific epitopes and ChTR1. Specific antibody production was determined by ELISA (B23) . For each immunization, representative results of three individual *mice* are shown...

...0/5 -

TCCATGAGCTTCCTGAT

GCT

CpG 3: None 0/5 -

TCCATGACGTTCCTGAC

GTT

End Table: Columns 1 - 4 of 4

Figure F3

Caption: Blood vessels in *mice* immunized with C. trachomatis 60-kD CRP-derived peptide (B10) (B30) . (A) Thickening of the arterial wall and perivascular fibrotic changes in *mice* immunized with ChTR1. The perivascular mononuclear inflammatory cells are apparent. (B) Normal morphology of the cardiac artery in *mice* immunized with FCA alone. (C) Occlusion of cardiac blood vessels in *mice* immunized with ChTR1. (D) No occlusions in cardiac blood vessels were seen in control *mice* immunized with FCA alone. (A and B) Elastica staining for collagen (red) to detect fibrotic changes. (C and D) H&E staining. Magnification, x 320...

...Figure Removed

Figure F4

Caption: Serum IgG antibody production in C. trachomatis-infected *mice*. Eight-week-old female BALB/c *mice* were inoculated either intranasally or intravaginally with the indicated doses of C. trachomatis MoPn IFUs (B31) . Thirty-six days (intranasal infection) or 42 days (intravaginal infection) after the inoculation, serum was collected and specific IgG antibody

production was determined by ELISA (Fig. 2B). Representative data from individual *mice* are shown...

References and Notes:

...in FCA (1 mg/ml) and emulsified in a 1:1 dilution with phosphate-buffered saline, were injected twice into 6-week-old BALB/c *mice* (50 (mu) g of peptide per mouse) as described (B8...

...17. BALB/c *mice* were immunized twice with ChTR1 in FCA (B10) , and hearts were removed 21 days after the initial immunization. Immunoperoxidase staining of histological heart sections was...20. Spleens from M7Aa-, ChTR1-, or kka-immunized BALB/c *mice* (B10) were removed 21 days after the first immunization, and T cells were enriched by negatively sorting out CD11b-, Gr1-, and B220-expressing cells with...

...21. For adoptive transfer of ChTR1 peptide-induced inflammatory heart disease into nonimmunized recipient *mice*, 6-week-old donor BALB/c *mice* were immunized twice with ChTR1 peptide in FCA or with FCA only (B10) . Twenty-one days after the initial immunization, splenic T cells (B20) were...

...syngeneic splenocytes pulsed with ChTR1 peptide (50 (mu) g/ml) for 4 days in the presence of mrIL-2 (50 U/ml). Recipient BALB/c *mice* were injected intraperitoneally with lipopolysaccharide (25 (mu) g per mouse) on days 0 and 4, and 1 x 10.sup(6) in vitro-stimulated cells from immunized donor *mice* were injected intravenously on day 7 (B7) . Transferred donor T cells (>95%) had a TCRa (beta) .sup(+)CD4.sup(+) CD69.sup(+)CD25.sup(+) phenotype...oligodeoxynucleotides (ODNs) were derived either from C. trachomatis DNA (CpG 1) or from previously reported bacterial DNA sequences (CpGs 2 and 3) (B24) . ODNs were *phosphorothioate* modified to increase their in vivo stability. ODNs (30 (mu) g in 100 (mu) l of 0.15 mM NaCl buffer) were administered ip at the time of the immunizations. BALB/c *mice* were subcutaneously immunized twice at a 7-day interval with the M7Aa peptide (50 (mu) g per mouse) in a 1:1 emulsion with mineral...as described [S. Pal, I. Theodor, E. M. Peterson, L. M. de la Maza, Infect. Immun. 65, 3361 (1997)]. Eight-week-old female BALB/c *mice* were inoculated either intranasally with 0 (sham), 1 x 10.sup(3), 1 x 10.sup(4), 1 x 10.sup(5), or 1 x ...

?

Set	Items	Description
S1	2069	CPG (S) ANTIGEN?
S2	312	S1 (S) ADMINIST?
S3	85	RD (unique items)
S4	67	S3 AND (MOUSE OR MICE)
S5	0	S3 AND (IN VIVO)
S6	24	S3 (S) (SUBSEQUENT? OR FOLLOW?)

>>>KWIC option is not available in file(s): 41, 77, 399

6/3,K/1 (Item 1 from file: 5)

13657441 BIOSIS NO.: 200200286262

Plasmid vectors encoding cholera toxin or the heat-labile enterotoxin from Escherichia coli are strong adjuvants for DNA vaccines.

AUTHOR: Arrington Joshua; Braun Ralph P; Dong Lichun; Fuller Deborah H; Macklin Michael D; Umlauf Scott W; Wagner Sarah J; Wu Mary S; Payne Lendon G; Haynes Joel R(a)
AUTHOR ADDRESS: (a)PowderJect Vaccines, Inc., 585 Science Dr., Madison, WI, 53711**USA E-Mail: joelhaynes@powderject.com

JOURNAL: Journal of Virology 76 (9):p4536-4546 May, 2002
MEDIUM: print
ISSN: 0022-538X
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

...ABSTRACT: B subunits of the Escherichia coli heat-labile enterotoxin (LT) were evaluated for their ability to serve as genetic adjuvants for particle-mediated DNA vaccines *administered* to the epidermis of laboratory animals. Both the CT and the LT vectors strongly augmented Th1 cytokine responses (gamma interferon (IFN-gamma)) to multiple viral *antigens* when codelivered with DNA vaccines. In addition, Th2 cytokine responses (interleukin 4 (IL-4)) were also augmented by both sets of vectors, with the effects of the LT vectors on IL-4 responses being more *antigen* dependent. The activities of both sets of vectors on antibody responses were *antigen* dependent and ranged from no effect to sharp reductions in the immunoglobulin G1 (IgG1)-to-IgG2a ratios. Overall, the LT vectors exhibited stronger adjuvant effects...

...T-cell responses than did the CT vectors, and this was correlated with the induction of greater levels of cyclic AMP by the LT vectors *following* vector transfection into cultured cells. The adjuvant effects observed in vivo were due to the biological effects of the encoded proteins and not due to *CpG* motifs in the bacterial genes. Interestingly, the individual LT A and B subunit vectors exhibited partial adjuvant activity that was strongly influenced by the presence...

6/3,K/2 (Item 2 from file: 5)

13654130 BIOSIS NO.: 200200282951

Parenteral and mucosal prime-boost immunization strategies in mice with hepatitis B surface antigen and CpG DNA.

AUTHOR: McCluskie Michael J(a); Weeratna Risini D; Payette Paul J; Davis Heather L
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JOURNAL: FEMS Immunology and Medical Microbiology 32 (3):p179-185 18 February, 2002
MEDIUM: print
ISSN: 0928-8244
DOCUMENT TYPE: Article
RECORD TYPE: Abstract

LANGUAGE: English

ABSTRACT: Synthetic oligodeoxynucleotides (ODN) containing immunostimulatory *CpG* motifs (*CpG* ODN) are potent adjuvants to protein *antigens* *administered* by parenteral or mucosal routes to BALB/c mice. To date, there have been no studies using combined parenteral/mucosal approaches with *CpG* DNA as adjuvant. In this study we evaluated different parenteral prime-mucosal boost and mucosal prime-parenteral boost strategies using hepatitis B surface *antigen* (HBsAg) alone or with different adjuvants: aluminum hydroxide (alum), cholera toxin (CT), *CpG* ODN. In addition, since *CpG* ODN has previously been shown to act synergistically with other adjuvants after parenteral or mucosal delivery, we also evaluated adjuvant combinations: alum+*CpG* ODN and CT+*CpG* ODN. The effects of adjuvant and *administration* strategy on systemic and mucosal humoral responses were measured, as well as cell-mediated immune responses (cytotoxic T lymphocyte activity). These results were compared to...

...all be significantly enhanced by mucosal boosting and despite the fact that intramuscular immunization alone could not induce mucosal IgA, it could prime for a *subsequent* mucosal boost. In addition, the presence of adjuvant at time of boosting could influence the nature of *subsequent* immune responses (Th1 vs. Th2). Mice primed intranasally could have their systemic immune responses boosted with a parenteral *administration* and it was also possible to enhance mucosal responses induced by intranasal prime with an intramuscular boost.

6/3,K/3 (Item 3 from file: 5)

13620548 BIOSIS NO.: 200200249369

Yersinia enterocolitica as a vehicle for a naked DNA vaccine encoding Brucella abortus bacterioferritin or P39 antigen.

AUTHOR: Al-Mariri Ayman; Tibor Anne; Lestrate Pascal; Mertens Pascal; De Bolle Xavier; Letesson Jean-Jacques(a)
AUTHOR ADDRESS: (a)Unite de Recherche en Biologie Moleculaire (URBM), Laboratoire d'Immunologie et de Microbiologie, Facultes Universitaires Notre-Dame de la Paix, Rue de Bruxelles 61, B-5000, Namur**Belgium
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JOURNAL: Infection and Immunity 70 (4):p1915-1923 April, 2002
MEDIUM: print
ISSN: 0019-9567
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

...ABSTRACT: and humans. The protective immune response against Brucella involves both humoral and cell-mediated immunity. In previous studies, we demonstrated that the T-dominant Brucella *antigens* bacterioferritin (BFR) and P39 *administered* either as *CpG* adjuvant recombinant proteins or as naked-DNA plasmids induced a specific Th1-biased immune response in mice. In order to improve the protection conferred by...

...antibodies, we used live attenuated Yersinia enterocolitica serotypes O:3 and O:9 as delivery vectors for naked-DNA plasmids encoding these BFR and P39 *antigens*. *Following* two intragastric immunizations in BALB/c mice, the Yersinia vectors harboring a DNA vaccine encoding BFR or P39 induced *antigen*-specific serum immunoglobulin and Th1-type responses (both lymphocyte proliferation and gamma interferon production) among splenocytes. Moreover, as expected, antibodies recognizing Brucella abortus 544 lipopolysaccharide...

...LPS antibodies in protection. The best protection was conferred by a serotype O:9 strain carrying pCIP39, confirming the importance of the P39 T-cell *antigen* in this mechanism.

6/3,K/4 (Item 4 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13591728 BIOSIS NO.: 200200220549
Endothelial cell-derived growth factor expands murine hematopoietic progenitor cells and DC precursor cells in vivo and increases the protective response to autologous tumor vaccination.
AUTHOR: McCormick Alison A(a); Davis Thomas(a); Wannberg Sharon(a); Tuse Daniel(a)
AUTHOR ADDRESS: (a)Large Scale Biology, Corp., Vacaville, CA**USA
JOURNAL: Blood 98 (11 Part 1):p701a November 16, 2001
MEDIUM: print
CONFERENCE/MEETING: 43rd Annual Meeting of the American Society of Hematology, Part 1 Orlando, Florida, USA December 07-11, 2001
ISSN: 0006-4971
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Dendritic cells (DC) are potent *antigen* processing and presenting cells considered to be essential for initiating rapid and efficient immune responses, and possess the unique ability to stimulate naive T-cells and B-cells. Increasing vaccine potency by stimulating *antigen* uptake and presentation by DC has become a major area of research in the past 10 years. We have found that treating mice with porcine...

...for 7 consecutive days. Then, vaccine groups received 15 mug of protein derived from the 38C13 mouse B-cell lymphoma (a tumor-associated syngeneic self-*antigen* protein), s.c. at 2-week intervals for a total of two vaccinations. To ensure activation of DC at the site of vaccine injection, we mixed the vaccine with either control vehicle or 10 mug of *cpG* DNA oligomer (Hartman, et al., PNAS 1999, 96(16): 9305-10). Ten days after each vaccination, humoral anti-idiotypic immmunoglobulin levels were determined by ELISA. A pronounced anti-38C13 immune response was detected as early as 10 days *following* the first vaccination compared to control groups which had little or no detectable response. Isotype analysis revealed a predominantly IgG2a response after a single vaccination, suggesting early and robust Th1-type B-cell help. After two vaccinations, mice treated with EDHGF+vaccine, or vaccine alone in the absence of *cpG* immunization, had significantly lower serum anti-38C13 titers with little IgG2a isotype. Two weeks after the second vaccination, animals were challenged with a lethal dose of *antigen*-expressing 38C13 lymphoma tumor cells. Animals pre-treated with EDHGF *followed* by vaccine+*CpG* DNA vaccination had significantly better survival than controls, vaccine treatment alone, or vaccine+*cpG*. These results suggest that in vivo expansion of DC precursors through *administration* of EDHGF augments vaccine potency by increasing *antigen* uptake and *antigen* presentation. Our results represent an important strategy for increasing the effectiveness of vaccination without modification of the *antigen* and without purification of DC.

6/3,K/5 (Item 5 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13242294 BIOSIS NO.: 200100449443
Serum and mucosal immune responses to an inactivated influenza virus vaccine induced by epidermal powder immunization.
AUTHOR: Chen Dexiang(a); Periwal Sangeeta B; Larrivee Katherine; Zuleger Cindy; Erickson Cherie A; Endres Ryan L; Payne Lendon G
AUTHOR ADDRESS: (a)PowderJect Vaccines, Inc., 585 Science Dr., Madison, WI, 53711: dexiangchen@powderject.com**USA
JOURNAL: Journal of Virology 75 (17):p7956-7965 September, 2001
MEDIUM: print
ISSN: 0022-538X

DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: Both circulating and mucosal antibodies are considered important for protection against infection by influenza virus in humans and animals. However, current inactivated vaccines *administered* by intramuscular injection using a syringe and needle elicit primarily circulating antibodies. In this study, we report that epidermal powder immunization (EPI) via a unique powder delivery system elicits both serum and mucosal antibodies to an inactivated influenza virus vaccine. Serum antibody responses to influenza vaccine *following* EPI were enhanced by codelivery of cholera toxin (CT), a synthetic oligodeoxynucleotide containing immunostimulatory *CpG* motifs (*CpG* DNA), or the combination of these two adjuvants. In addition, secretory immunoglobulin A (sIgA) antibodies were detected in the saliva and mucosal lavages of the...

...titers. The local origin of the sIgA antibodies was further shown by measuring antibodies released from cultured tracheal and small intestinal fragments and by detecting *antigen*-specific IgA-secreting cells in the lamina propria using ELISPOT assays. EPI with a single dose of influenza vaccine containing CT or CT and *CpG* DNA conferred complete protection against lethal challenges with an influenza virus isolated 30 years ago, whereas a prime and boost immunizations were required for protection...

...the absence of an adjuvant. The ability to elicit augmented circulating antibody and mucosal antibody responses makes EPI a promising alternative to needle injection for *administering* vaccines against influenza and other diseases.

6/3,K/6 (Item 6 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

13060550 BIOSIS NO.: 200100267699
CpG containing DNA promotes Th1 responses, but fails to sustain long-lasting T cell memory.
AUTHOR: Tian Jide(a); Olcott Angelica(a); Lu Yuxin(a); Hanssen Lori(a); Zekzer Dan(a); Ausubel Lara(a); Ornelas Richard(a); Melamed Esther(a); Quach Phung(a); Chaaban Manar(a); Kaufman Daniel(a)
AUTHOR ADDRESS: (a)University of California Los Angeles, 10833 Le Conte Ave., Los Angeles, CA, 90095**USA
JOURNAL: FASEB Journal 15 (4):pA652 March 7, 2001
MEDIUM: print
CONFERENCE/MEETING: Annual Meeting of the Federation of American Societies for Experimental Biology on Experimental Biology 2001 Orlando, Florida, USA March 31-April 04, 2001
ISSN: 0892-6638
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: Bacterial DNA containing *CpG* motifs has been shown to have strong adjuvanicity, and may help to provide a new generation of vaccines. However, little is known about the quality and quantity of long-term T cell memory responses, which are primed by *antigen*/DNA. Here, we examined the effects of *antigen*/DNA co-*administration* on T cell immunity in both Th1-biased and Th2-biased mice. Immunization of mice with *antigen* plus bacterial (plasmid pUC18), but not vertebrate DNA, primed a high frequency of Th1 cells and promoted the development of DTH responses. Unexpectedly, the *antigen*/pUC18 primed Th1 responses declined rapidly after immunization and virtually disappeared after 8 weeks. In contrast, *antigen*/CFA primed T cells sustained a comparable level even at 12 weeks post immunization. Moreover, mice which had been presensitized with *antigen*/pUC18 failed to display a memory T cell response *following* rechallenge with *antigen*/CFA, and rather only

displayed a primary T cell response. Together, these data suggest that *CpG* DNA can act as an adjuvant to prime Th1 responses, but fails to sustain long term T cell memory. Our findings provide new insights into *CpG*-DNA adjuvanicity and may aid in the design of future vaccination strategies for infectious diseases and cancer.

6/3,K/7 (Item 7 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

13007520 BIOSIS NO.: 200100214669

Adjuvantation of epidermal powder immunization.

AUTHOR: Chen Dexiang(a); Erickson Cherie A; Endres Ryan L; Periwal Sangeeta B; Chu Qili; Shu Cassandra; Maa Yuh-Fun; Payne Lendon G
AUTHOR ADDRESS: (a)PowderJect Vaccines Inc., 585 Science Drive, Madison, WI, 53711: dexiangchen@powderject.com**USA
JOURNAL: Vaccine 19 (20-22):p2908-2917 6 April, 2001
MEDIUM: print
ISSN: 0264-410X
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RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: The skin is an immunologically active site and an attractive vaccination route. All current vaccines, however, are *administered* either orally, intramuscularly, or subcutaneously. We previously reported that epidermal powder immunization (EPI) with an extremely small dose of powdered influenza vaccine induces protective immunity in mice. In this study, we report that commonly used adjuvants can be used in EPI to further enhance the immune responses to an *antigen*. The IgG antibody response to diphtheria toxoid (DT) *following* EPI was augmented by 25- and 250-fold, when 1 mug DT was co-delivered with aluminum phosphate (alum) and a synthetic oligonucleotide containing *CpG* DNA motifs (*CpG* DNA), respectively. These antibodies had toxin-neutralization activity and were long lasting. Furthermore, EPI using an adjuvant selectively activated different subsets of T helper cells...

...predominantly IgG1 subclass antibody response and elevated level of IL-4 secreting cells. These are indicative of Th2-type immunity. In contrast, co-delivery of *CpG* DNA adjuvant via EPI led to Th-1 type of response as characterized by the increased production of IgG2a antibodies and IFN-gamma secreting cells...

...using appropriate adjuvants can produce an augmented antibody response and desirable cellular immune responses. EPI is a promising immunization method that may be used to *administer* a broad range of vaccines including vaccines with adjuvants.

6/3,K/8 (Item 8 from file: 5)

DIALOG(R)File 5:Biosis Previews(R)

12974712 BIOSIS NO.: 200100181861

Intratracheal *administration* of *CpG*-ODN with or *following* *antigen* challenge inhibits Th2-mediated airway hypersensitivity in a mouse model of allergic asthma.

AUTHOR: Teper Ariel; Schofield Brian; Kattan Meyer(a); Sampson Hugh A(a); Li Xiu-Min
AUTHOR ADDRESS: (a)Mount Sinai School of Medicine, New York, NY**USA
JOURNAL: Journal of Allergy and Clinical Immunology 107 (2):pS99 February, 2001
MEDIUM: print
CONFERENCE/MEETING: 57th Annual Meeting of the American Academy of Allergy, Asthma and Immunology New Orleans, Louisiana, USA March 16-21, 2001
ISSN: 0091-6749

RECORD TYPE: Citation
LANGUAGE: English
SUMMARY LANGUAGE: English

Intratracheal *administration* of *CpG*-ODN with or *following* *antigen* challenge inhibits Th2-mediated airway hypersensitivity in a mouse model of allergic asthma.

6/3,K/9 (Item 9 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

12908183 BIOSIS NO.: 200100115332
CpG oligodeoxynucleotide vaccination suppresses IgE induction but may fail to down-regulate ongoing IgE responses in mice.
AUTHOR: Peng Zhikang(a); Wang Hongsheng; Mao Xiaojuan; HayGlass Kent T; Simons F Estelle R
AUTHOR ADDRESS: (a)University of Manitoba, 715 McDermot Avenue, 532 John Buhler Research Centre, Winnipeg, Manitoba, R3E 3P5**Canada
JOURNAL: International Immunology 13 (1):p3-11 January, 2001
MEDIUM: print
ISSN: 0953-8178
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: *Antigen*-specific IgE plays an important role in the pathogenesis of allergic disorders. Immunostimulatory *CpG* motifs (*CpG*) in bacterial DNA or synthesized oligodeoxynucleotides (ODN) are gaining recognition as potential immunomodulators for switching on protective Th1-mediated immunity and preventing or potentially inhibiting Th2-dependent allergic responses. To date, allergic models used in *CpG* ODN studies have been established by immunization of mice with allergen in the presence of adjuvant. This, in addition to failure to assess specific IgE production in most of the studies, has limited understanding of the role of *CpG* ODN vaccination in allergic responses. Here, we examine the effects of synthesized *CpG* ODN on both developing and ongoing IgE responses in mice sensitized using a recombinant mosquito salivary *antigen* (rAed a 2) without adjuvant. Pretreatment of mice with *CpG* ODN mixed with rAed a 2 successfully inhibited *subsequent* induction of serum rAed a 2-specific IgE (but not IgG1) and *antigen* -induced IL-4 and IL-5 production in spleen cells. This was associated with an increase of serum IgG2a and IL-12, and increased IFN-gamma and IL-12 production by spleen cells. In this model, however, co-*administration* of *CpG* ODN with rAed a 2 to presensitized mice failed to down-regulate ongoing IgE responses despite significant up-regulation of serum IL-12 and specific IgG2a. Strikingly, a transient skin delayed-type hypersensitivity reaction occurred in *CpG* ODN-treated mice. These observations provide a new insight into the potential therapeutic application of *CpG* ODN to allergic disorders.

6/3,K/10 (Item 10 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

12896499 BIOSIS NO.: 200100103648
Human immunodeficiency virus (HIV)-specific immune responses are generated with the simultaneous vaccination of a gp120-depleted, whole-killed HIV-1 immunogen with cytosine-phosphorothioate-guanine dinucleotide immunostimulatory sequences of DNA.
AUTHOR: Moss Ronald B(a); Diveley Jocelyn; Jensen Fred C; Gouveia Erin; Carlo Dennis J
AUTHOR ADDRESS: (a)Immune Response Corporation, 5935 Darwin Court, Carlsbad, CA, 92008: shotdoc@imnr.com**USA
JOURNAL: Journal of Human Virology 4 (1):p39-43 January-February, 2001

MEDIUM: print
ISSN: 1090-9508
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

ABSTRACT: Objectives: We compared the effect of priming with a synthetic oligodeoxynucleotide (ODN) immunostimulatory DNA sequence *followed* by vaccination with human immunodeficiency virus type 1 (HIV-1) in incomplete Freund's adjuvant (IFA) or HIV-1 *antigen* alone to the simultaneous *administration* of immunostimulatory sequences (ISS) with HIV-1 in IFA. Methods: We examined immune function involving interferon-gamma (IFN-gamma) production, RANTES (regulated upon activation, normal...

...production, and lymphocyte proliferation, all of which appear to be augmented in HIV-1-exposed, but uninfected, individuals. Results: We demonstrate that similar levels of *antigen*-specific IFN-gamma were produced from lymph node cells of the animals immunized with HIV-1 *antigen* in IFA containing the *CpG* ODN 1826 (ISS; mean +- SE = 450.8 +- 224.3 pg/mL) and the group of animals primed with the ODN before injection with the HIV-1 in IFA (mean +- SE = 377.7 +- 294.8 pg/mL) or HIV-1 *antigen* alone (IFN-gamma = 0 pg/mL). However, the group that received the HIV-1 in IFA plus ISS mounted a stronger lymphocyte proliferation (mean net...

...plus ISS also showed a higher p24-specific response that was predominantly of the immunoglobulin G IgG2b isotype. Conclusion: These results suggest that the simultaneous *administration* of the ISS in the HIV-1 in IFA emulsion may be a condidate for testing in non-human primates and in human studies as...

6/3,K/11 (Item 11 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

12279232 BIOSIS NO.: 200000032734
LPD lipopolyplex initiates a potent cytokine response and inhibits tumor growth.
AUTHOR: Whitmore M; Li S; Huang L(a)
AUTHOR ADDRESS: (a)Laboratory of Drug Targeting, Department of Pharmacology, University of Pittsburgh School of Medicine, W1351 Biomedical Sciences Tower, Pittsburgh, PA, 15261**USA
JOURNAL: Gene Therapy 6 (11):p1867-1875 Nov., 1999
ISSN: 0969-7128
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English
SUMMARY LANGUAGE: English

...ABSTRACT: has recently developed a lipopolyplex consisting of DOTAP:cholesterol liposomes, protamine sulfate, and plasmid DNA (LPD) that provides improved systemic gene delivery compared with lipoplex *following* tail vein injection in mice. Because endothelial cells are the primary cells transfected in the lung, it was hypothesized that LPD might be an effective...
...of cytokine were observed in all organs (lung, liver, kidney and spleen) where LPD is known to have gene transfer activity. Methylation of immune-stimulatory *CpG* motifs in the plasmid component of LPD inhibited the proinflammatory cytokine response as well as the antitumor effect of LPD in both tumor systems. This suggests that i.v. *administration* of LPD elicits a systemic proinflammatory cytokine response that mediates the antitumor activity of the lipopolyplex. In addition, the antitumor activity was not observed in...

...mice suggesting a possible role for B or T lymphocytes in the antitumor

response initiated by LPD. This represents the first demonstration that an intravenously *administered* cationic liposome-based nonviral vector can promote a systemic, Th1-like innate immune response. The immune adjuvant properties of LPD might prove to be suitable for delivering tumor-specific *antigens* in the context of DNA vaccination.

6/3,K/12 (Item 12 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

10895472 BIOSIS NO.: 199799516617
Crystal structure of carboxypeptidase G-2, a bacterial enzyme with applications in cancer therapy.
AUTHOR: Rowsell Sian; Paupit Richard A; Tucker Alec D; Melton Roger G; Blow David M; Brick Peter(a)
AUTHOR ADDRESS: (a)Blackett Lab., Imperial Coll., London SW7 2BZ**UK
JOURNAL: Structure (London) 5 (3):p337-347 1997
ISSN: 0969-2126
RECORD TYPE: Abstract
LANGUAGE: English

...ABSTRACT: Carboxypeptidase G enzymes hydrolyze the C-terminal glutamate moiety from folic acid and its analogues, such as methotrexate. The enzyme studied here, carboxypeptidase G-2 (*CPG*-2), is a dimeric zinc-dependent exopeptidase produced by Pseudomonas sp. strain RS-16. *CPG*-2 has applications in cancer therapy: *following* its *administration* as an immunoconjugate, in which *CPG*-2 is linked to an antibody to a tumour-specific *antigen*, it can enzymatically convert *subsequently* *administered* inactive prodrugs to cytotoxic drugs selectively at the tumour site. *CPG*-2 has no significant amino acid sequence homology with proteins of known structure. Hence, structure determination of *CPG*-2 was undertaken to identify active-site residues, which may in turn provide ideas for protein and/or substrate modification with a view to improving its therapeutic usefulness. Results: We have determined the crystal structure of *CPG*-2 at 2.5 ANG resolution using multiple isomorphous replacement methods and non-crystallographic symmetry averaging. Each subunit of the molecular dimer consists of a...

...3 ANG apart. This distance is bridged by two shared zinc ligands, an aspartic acid residue and a hydroxyl ion. Conclusions: We find that the *CPG*-2 catalytic domain has structural homology with other zinc-dependent exopeptidases, both those with a single zinc ion and those with a pair of zinc...

6/3,K/13 (Item 13 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

10566916 BIOSIS NO.: 199699188061
In vitro and in vivo characterisation of a recombinant carboxypeptidase G-2::anti-CEA scFv fusion protein.
AUTHOR: Michael N Paul(a); Chester Kerry A; Melton Roger G; Robson Lynda; Nicholas William; Boden Joan A; Pedley R Barbara; Begent Richard H J; Sherwood Roger F; Minton Nigel P
AUTHOR ADDRESS: (a)Dep. Molecular Microbiology, Res. Div., Centre Applied Microbiology Res., Salisbury, Wiltshire S**UK
JOURNAL: Immunotechnology (Amsterdam) 2 (1):p47-57 1996
ISSN: 1380-2933
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

...ABSTRACT: tumour. After allowing sufficient time for the conjugate to localise at the turnout and clear from the circulatory system, a relatively non-toxic prodrug is *administered*. This prodrug is converted to a highly cytotoxic drug by the action of the targeted enzyme localised

at the tumour site. Objectives: To construct gene fusions between the pseudomonad carboxypeptidase G-2 (*CPG*-2) gene and DNA encoding MFE-23 (an anti-carcinoembryonic *antigen* (CEA) single-chain Fv (scFv) molecule), derived from a phage display library. To overexpress the resultant gene fusions in Escherichia coli, and assess the in vitro and in vivo properties of the purified fusion proteins. Study design: To introduce unique cloning restriction sites into the 5'-end of the *CPG*-2 gene by site-directed mutagenesis to facilitate fusion to the 3'-end of the gene encoding MFE-23 (constructs with or without a flexible...

...to direct the fusion proteins produced to the periplasmic space of E. coli through translational coupling to the pelB signal peptide. Results: Biologically active recombinant *CPG*-2::MFE-23 scFv fusion proteins were produced in E. coli and shown to possess enzyme and anti-CEA activity. Affinity chromatography *followed* by size exclusion gel filtration yielded approximately 0.7-1.4 mg/l from shake flask culture. The fusion protein in which the enzyme and...

6/3,K/14 (Item 14 from file: 5)
DIALOG(R)File 5:Biosis Previews(R)

08902379 BIOSIS NO.: 199396053880
Tumour necrosis factor increases tumour uptake of co-administered antibody-carboxypeptidase G-2 conjugate.
AUTHOR: Melton R G(a); Rowland J A; Pietersz G A; Sherwood R F; McKenzie I F C
AUTHOR ADDRESS: (a)Div. Biotechnol., PHLS Centre Applied Microbiol. and Res., Porton Down, Salibury, Wilts SP4 0JG**UK
JOURNAL: European Journal of Cancer 29A (8):p1177-1183 1993
ISSN: 0959-8049
DOCUMENT TYPE: Article
RECORD TYPE: Abstract
LANGUAGE: English

ABSTRACT: Increased tumour uptake of antibodies and antibody-drug conjugates has been demonstrated *following* pretreatment of animals with recombinant human tumour necrosis factor-alpha (rTNF-alpha) and interleukin 2 immunoconjugates. The experiments reported here were performed to determine whether...

...thymoma E3 were simultaneously injected with 2.0 mu-grTNF-alpha and 3.5 mu-g(74 kBq) 125I-labelled murine anti-Ly-2.1-*CPG*-2 conjugate. Mice in control groups received phosphate buffered saline in place of rTNF-alpha. The conjugated corresponded in molecular weight to a mixture of 1:1 and 2:1 (*CPG*-2:IgG) conjugate and retained its *antigen* binding specificity and enzymic activity in vitro. A significant increase in tumour uptake was observed 24 h after *administration* when rTNF-alpha-treated animals were compared to controls (28.1 +- 9.7%/g and 11.6 +- 2.3%/g, respectively). Other tissues, most notably...

6/3,K/15 (Item 1 from file: 34)
DIALOG(R)File 34:SciSearch(R) Cited Ref Sci

08022716 Genuine Article#: 237TM No. References: 267
Title: Novel strategies using DNA for the induction of mucosal immunity
Author(s): McCluskie MJ; Davis HL (REPRINT)
Corporate Source: OTTAWA HOSP,LOEB HLTH RES INST, 725 PARKDALE AVE/OTTAWA/ON K1Y 4E9/CANADA/ (REPRINT); OTTAWA HOSP,LOEB HLTH RES INST/OTTAWA/ON K1Y 4E9/CANADA/; UNIV OTTAWA,FAC HLTH SCI, SCH REHABIL SCI/OTTAWA/ON K1N 6N5/CANADA/; UNIV OTTAWA,FAC MED, DEPT BIOCHEM MICROBIOL & IMMUNOL/OTTAWA/ON K1N 6N5/CANADA/; UNIV OTTAWA,FAC MED, DEPT CELLULAR & MOL MED/OTTAWA/ON K1N 6N5/CANADA/; CPG IMMUNOPHARMACEUT INC,/WELLESLEY//MA/
Journal: CRITICAL REVIEWS IN IMMUNOLOGY, 1999, V19, N4, P303-329

ISSN: 1040-8401 Publication date: 19990000
Publisher: BEGELL HOUSE INC, 79 MADISON AVE, SUITE 1205, NEW YORK, NY 10016-7892
Language: English Document Type: REVIEW (ABSTRACT AVAILABLE)

Abstract: The mucosal surfaces are the primary sites for transmission of most infectious diseases. However, most conventional vaccines are *administered* parenterally [e.g., by intramuscular (IM) or intradermal (ID) injection] and induce systemic but rarely mucosal immunity. Novel vaccination strategies capable of inducing both systemic...
...and morbidity worldwide. One of the most exciting advances in vaccine technology in recent years has been the development of DNA vaccines, through which the *antigen* is synthesized in viva after direct introduction of its encoding sequences. The vast majority of DNA vaccines have been delivered parenterally; however, in recent years a number of studies have reported successful mucosal immunization with DNA vaccines. The induction of strong immune responses *following* the introduction of DNA appears to be partly due to the potent adjuvant effect of unmethylated immunostimulatory *CpG* motifs present in the DNA backbone. Synthetic oligodeoxynucleotides (ODN) containing such immunostimulatory *CpG* motifs are potent adjuvants systemically and mucosally in mice, and have synergistic action with other adjuvants, such as alum and cholera toxin (CT). This article highlights the recent advances in vaccination strategies using DNA delivered to mucosal surfaces either as an *antigen*-encoding plasmid or as an adjuvant.

6/3,K/16 (Item 1 from file: 76)
DIALOG(R)File 76:Life Sciences Collection

02706289 5359393
Chemokines: Agents for the immunotherapy of cancer?
Homey, B.; Mueller, A.; Zlotnik, A.
Nature Reviews: Immunology vol. 2, no. 3, pp. 175-184 (2002)
ISSN: 1474-1733
DOCUMENT TYPE: Journal article; Review article LANGUAGE: ENGLISH
SUBFILE: Immunology Abstracts

...3b), CCL20 (MIP-3a), CCL21 (6Ckine), CXCL10 (interferon- gamma inducible protein-10, IP-10) and XCL1 (lymphotactin) alone can induce tumour regression and immunity to *subsequent* tumour challenge. Chemokines alone seem to show limited antitumour efficacy. However, new approaches are being developed that combine a chemoattractant (e.g. CCL19, CCL21, CXCL9...

...IL-12, granulocyte-macrophage colony-stimulating factor (GM-CSF)), which are known for their stimulating properties on T cells, natural killer (NK) cells or tumour *antigen*-pulsed dendritic cells (DCs). Chemokines might act as potent natural adjuvants for experimental antitumour immunotherapy. Their combination with tumour peptide-pulsed DCs and direct coupling to tumour *antigen* or immunostimulatory cytokines results in synergistic antitumour activity. This is a way of reducing toxic side effects. The combination of tumour *antigen*-releasing therapies (chemotherapy and radiation therapy) with chemokine delivery to sites of tumour *antigen* exposure, and the in vivo *administration* of `DC-poietins' such as FTL3 ligand or GM-CSF and other activation molecules (IL-2, IL-12, CD40L and *CpG*), offers promising strategies to induce strong and long-lived antitumour immunity. However, side effects such as the induction of autoimmunity and the concept of leukocyte...

6/3,K/17 (Item 1 from file: 135)
DIALOG(R)File 135:NewsRx Weekly Reports

0000035275 (USE FORMAT 7 OR 9 FOR FULLTEXT)
"Ambivalent Effects of CpG Oligodeoxynucleotides on the Immunogenicity of Soluble and Particulate HIV Vaccine Components."

AIDS Weekly, August 16, 1999, p.19

DOCUMENT TYPE: Research News LANGUAGE: English
RECORD TYPE: FULLTEXT
WORD COUNT: 362

...TEXT: by the authors to the Fourth European Conference on Experimental AIDS Research, held June 18-21, 1999 in Tampere, Finland, "Bacterial DNA sequences containing unmethylated *CpG* motifs have recently been proposed to exhibit immunostimulatory effects on B-, T-, and NK cells, leading to the induction of humoral and cell-mediated immune responses. Here, we investigated the immunomodulatory effects of *CpG*-containing oligodeoxynucleotides (*CpG*-ODNs) to W soluble HIV-1 gp160 Env proteins and (ii) Pr55gag virus-like particles (VLPs) in the BALB/c mouse model. *Administration* of gp160 resulted in a typical T-helper-2 (Th-2) response with high titers of IgG1 isotypes but a weak IgG2a response. Cultured splenocytes...
...IgG2 isotype antibodies, without affecting titers of other isotypes. Furthermore, elevated quantities of IFN-g but no IL-5 were secreted from restimulated splenocytes. Thus, *CpG*-ODNs were useful as an adjuvant to induce a typical Th-0/Th-1 response to soluble protein *antigens*. However, no specific CTL response was observed *following* an-immunization with gp160 in absence and presence of *CpG*-ODNS. In contrast to soluble gp160, immunization with chimeric Pr55gag VLPS stimulated immune responses with a Th-0/Th-1 bias, with the expansion of...

...specific CD4+ T cells producing high quantities of IFN-g, the induction of high titers of both IgG1 and IgG2a isotypes and the priming of *antigen* -specific cytotoxic T cells. Coadministration of Pr55gag VLPs and *CpG* -ODNs had little if any effect on the anti-Gag isotype responses. However, IFN-g secretion of specifically restimulated splenocytes from mice, immunized with a VLP/*CpG*-ODN combination, was 50-fold decreased. Furthermore, the induction of cytotoxic T cells was significantly impaired by the coadministration of *CpG*-ODNs. These results strongly indicate an ambivalent effect of *CpG*-ODNs on the immunological properties of soluble (gp 160) and particulate (Pr55gagALPs) vaccine preparations." (Authors) D. Ludwig, R. Schirmbeck, H. Wolf, J. Reimann and R...

6/3,K/18 (Item 1 from file: 144)
DIALOG(R)File 144:Pascal

15104071 PASCAL No.: 01-0264445
Adjuvantation of epidermal powder immunization
DEXIANG CHEN; ERICKSON Cheric A; ENDRES Ryan L; PERIWAL Sangeeta B; QILI CHU; SHU Cassandra; MAA Yuh-Fun; PAYNE Lendon G
PowderJect Vaccines Inc., 585 Science Drive, Madison, WI 53711, United States; PowderJect Technologies Inc., 6511 Dumbarton Circle, Fremont, CA, United States
Journal: Vaccine, 2001, 19 (20-22) 2908-2917
Language: English

The skin is an immunologically active site and an attractive vaccination route. All current vaccines, however, are *administered* either orally, intramuscularly, or subcutaneously. We previously reported that epidermal powder immunization (EPI) with an extremely small dose of powdered influenza vaccine induces protective immunity in mice. In this study, we report that commonly used adjuvants can be used in EPI to further enhance the immune responses to an *antigen* . The IgG antibody response to diphtheria toxoid (DT) *following* EPI was augmented by 25- and 250-fold, when 1 mu g DT was co-delivered with aluminum phosphate (alum) and a synthetic oligonucleotide containing *CpG* DNA motifs (*CpG* DNA), respectively. These antibodies had toxin-neutralization activity and were long lasting. Furthermore, EPI using an adjuvant selectively activated different subsets of T helper cells...

...predominantly IgGI subclass antibody response and elevated level of IL-4 secreting cells. These are indicative of Th2-type immunity. In contrast, co-delivery of *CpG* DNA adjuvant via EPI led to Th-1 type of response as characterized by the increased production of IgG2a antibodies and IFN-gamma secreting cells...

... using appropriate adjuvants can produce an augmented antibody response and desirable cellular immune responses. EPI is a promising immunization method that may be used to *administer* a broad range of vaccines including vaccines with adjuvants.

6/3,K/19 (Item 1 from file: 266)
DIALOG(R)File 266:FEDRIP

00357563
IDENTIFYING NO.: 1Z01SC06526-11 AGENCY CODE: CRISP
MODULATION OF CELL GROWTH BY ANTISENSE AND ANTIGENE REAGENTS
PRINCIPAL INVESTIGATOR: NECKERS, LEONARD
ADDRESS: NCI SC, NIH
SPONSORING ORG.: DIVISION OF CLINICAL SCIENCES - NCI
FY : 2001

...SUMMARY: focus of this project is three-fold: (1) to characterize uptake and intra- cellular processing of unmodified and modified oligonucleotides; (2) to utilize antisense and *antigene* technology in several in vitro model systems to identify critical elements in cell proliferation/viral replication; and (3) to study the efficacy of antisense and *antigene* reagents as in vivo modulators of gene express- ion. (1) We have characterized the uptake of modified oligos as an energy-depend- ent, endocytic process...

... abl tyrosine kinase and other tyrosine kinases via direct interaction with the proteins. (5) We have been able to demonstrate significant prolongation of animal survival *following* injection of tumor cells treated with antisense to c-myc. At the same time such antisense treatment has no effect on normal bone marrow cells...

... ribozymes are delivered intracranially via osmotic pump. Preliminary data suggest that a combination of anti-VEGF and anti-FGF ribozymes significantly prolongs animal survival if *administration* is begun at the same time as tumor cell inoculation (using a human glioblastoma, U87). Ongoing studies will determine the distribution of ribozymes in brain...

... these ribozymes toward VEGF and FGF mRNA in the tumor cells. In additional animal studies, we have demonstrated significant anti-tumor activity of a unique *CpG* oligonucleotide. A single inoculation of mice with this oligonucleotide either pre- or post-tumor inoculation is able to prevent tumor growth, or cause tumor stasis...

6/3,K/20 (Item 2 from file: 266)
DIALOG(R)File 266:FEDRIP

00301519
IDENTIFYING NO.: 5R01CA85307-02 AGENCY CODE: CRISP
TUMOR RNA TRANSFECTED DENDRITIC CELL VACCINES
PRINCIPAL INVESTIGATOR: GILBOA, ELI
ADDRESS: DUKE UNIVERSITY MEDICAL CENTER BOX 2601 DURHAM, NC 27710
PERFORMING ORG.: DUKE UNIVERSITY, DURHAM, NORTH CAROLINA
SPONSORING ORG.: NATIONAL CANCER INSTITUTE
FY : 2001

...SUMMARY: strategies that will augment, in a significant manner, the therapeutic impact of this intervention modality. Animal models will be

used to (a) develop improved DC *antigen* presenting platforms and (b) design adjunct treatments to enhance, qualitatively as well as quantitatively, the immune response generated by the RNA transfected DC. In Specific...

... biological impact of various agents on DC maturation and then correlate with their potency to stimulate T cell responses and protective immunity. He will also *follow* up on preliminary observations suggesting that calreticulin (CRT) functions as a potent maturation agent. In Specific Aim 2, the applicant will test whether Flt-3L...
... the therapeutic benefit of this intervention modality. Methods to boost tumor-specific CD4+, as well as CD8+ T cell responses will be explored by co-*administration* of agents such as IL-2, IL-12, *CpG* ODNs, Poly I:C or anti-CTLA4 ab. Promising strategies indicated by the murine studies will be channeled into the applicant's ongoing clinical program...

6/3,K/21 (Item 3 from file: 266)
DIALOG(R)File 266:FEDRIP

00287226
IDENTIFYING NO.: 5P01AI28147-13 0001 AGENCY CODE: CRISP
MUCOSAL DNA AND PROTEIN VACCINES FOR AIDS PREVENTION
PRINCIPAL INVESTIGATOR: COMPANS, RICHARD W
ADDRESS: UNIVERSITY OF ALABAMA AT BIRMI UAB STATION BIRMINGHAM, AL 35294-2170
PERFORMING ORG.: UNIVERSITY OF ALABAMA AT BIRMINGHAM, BIRMINGHAM, ALABAMA
SPONSORING ORG.: NATIONAL INSTITUTE OF ALLERGY AND INFECTIOUS DISEASES
FY : 2001

...SUMMARY: sites. In this project, we will develop novel DNA vectors and delivery approaches to effectively prime the mucosal immune response, as well as particulate protein *antigens* which should be effective for boosting the resulting levels of mucosal immunity. We will construct DNA vectors which produce non-infectious VLPs which contain SIV Gag and HIV Env proteins (SHIV-89.6P VLPs). In addition, we will develop SHIV virus- like particles as protein *antigens* for use in boosting immune responses to DNA vaccines or poliovirus replicons. We will determine *antigens* for use in boosting immune responses to DNA vaccines or poliovirus replicons. We will determine humoral and cellular immune responses induced by SHIV-89.6P DNA vaccine and VLPs in preliminary studies in mice, in order to define the optimum protocol for *subsequent* use in rhesus macaques. The first specific aim is to develop more effective approaches for delivery of DNA expression vectors to mucosal surface. We have recently observed enhanced systemic and mucosal immune responses to DNA-encoded *antigens* after intranasal or oral delivery of DNA vectors when *administered* in a liposome preparation or when co-*administered* with a bioadhesive polymer. These approaches will be further developed by use of combinations of such polymers together with liposomes or other agents with known potential for enhancing DNA vaccine efficacy, such as *CpG* liposomes or other agents with known potential for enhancing DNA vaccine efficacy, such as *CpG* oligonucleotides. We will also determine the effects of co-*administration* of DNA plasmids encoding selected cytokines to mucosal surfaces. The second specific aim is to evaluate the use of virus-like particles (VLPs) for boosting...

...and mucosal immune responses to alternative types of immunogens in mice, and those approaches which are found to be most effective will be selected for *subsequent* studies in primates in projects 3 and 4.

6/3,K/22 (Item 1 from file: 357)
DIALOG(R)File 357:Derwent Biotech Res.

0252867 DBA Accession No.: 2000-07357
Plasmid DNA vaccines are effective in the absence of IFN-gamma - nucleic

acid vaccine used to induce humoral and cellular immune response independent of the presence of interferon-gamma

AUTHOR: Hassett D E; Zhang J; +Whitton J L
CORPORATE AFFILIATE: Scripps-Res.Inst.
CORPORATE SOURCE: Department of Neuropharmacology, CVN-9, The Scripps Research Institute, 10550 N. Torrey Pines Rd., La Jolla, CA 92037, USA. email:lwhitton@scripps.edu
JOURNAL: Virology (263, 1, 175-83) 1999
ISSN: 0042-6822 CODEN: VIRLAX
LANGUAGE: English

ABSTRACT: Injection of plasmid DNA derived from bacteria can be used to induce both humoral and cellular immune responses against *antigens* encoded by the plasmid. *CpG* sequences within the bacterium DNA are thought to enhance this process by stimulating secretion of proinflammatory cytokines, e.g. interferon-gamma (IGNg), by immune system cells. Despite evidence of IFNg being induced by *CpG* elements in plasmids, it remained unclear of the need for IFNg for successful immunization with nucleic acid vaccines. To investigate this, humoral and cellular immune...

...deficient mice inoculated with plasmid pCMVNP encoding the nucleoprotein of arena virus lymphocytic choriomeningitis virus (LCMV) were analyzed. Mice both with and without IFNg produced *antigen* specific CD8+ T-lymphocytes and virus-specific cytotoxic T-lymphocytes. Inoculation of either type of mice resulted in a significant reduction in virus titre *following* LCMV challenge, revealing IFNg is not required for successful nucleic acid vaccine *administration*. However IFNg-negative mice did show a difference in the IgG2a:IgG1 ratio, due to the need for IFNg to induce isotype switching. (43 ref)

6/3,K/23 (Item 1 from file: 370)

00500655 (USE 9 FOR FULLTEXT)

Pycnodysostosis, a Lysosomal Disease Caused by Cathepsin K Deficiency

Gelb, Bruce D.; Shi, Guo-Ping; Chapman, Harold A.; Desnick, Robert J.
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...Text: Bone resorption, a process mediated by osteoclasts, is characterized by the solubilization of inorganic mineral and *subsequent* proteolytic degradation of organic matrix, primarily type I collagen. In Pycno, osteoclast numbers are normal as are their ruffled borders and clear zones, but the...
...family by exon amplification from genomic DNA and by reverse transcriptase-polymerase chain reaction (RT-PCR) (B9) revealed a C-to-T transversion of a *CpG* dinucleotide at position 343 in the cDNA in both families, predicting an Arg.sup(113) --> Trp (R113W) substitution in the propeptide, near the putative cleavage...be heterozygous for markers across the Pycno critical region. Sequence analysis demonstrated heteroallelism for the G146R mutation and a C-to-T transition of a *CpG* dinucleotide at nucleotide 826, predicting an R241X nonsense mutation. Restriction analysis of amplified segments from genomic DNA with Bam I for G146R and Ava I...

...R241X mutation presumably will be null for cathepsin K activity. The

G146R mutation, found in the American Hispanic and Moroccan Arab families, occurred at a *CpG* dinucleotide and may prove to be a common mutation. Because this missense mutation would alter the charge of this residue, which resides near the active...

...K release by osteoclasts or other cells may be injurious. In these common disorders, down-regulation of gene expression by antisense RNA strategies or the *administration* of specific enzyme inhibitors may decrease pathologic bone resorption...the Israeli Arab family, is shown with the predicted elongation of the mature peptide by 19 residues. Abbreviations for the amino acid residues are as *follows*: A, Ala; D, Asp; F, Phe; G, Gly; H, His; I, Ile; K, Lys; L, Leu; P, Pro; Q, Gln; S, Ser; V, Val; and...

...polyclonal antibodies to human cathepsin K raised by injection of cathepsin K-maltose binding protein fusion protein into rabbits and purified by elution from immobilized *antigen* as described (B21) . Lane 1, nontransfected cells; lanes 2 and 3, 4 and 5, and 6 and 7, cells transfected with 3, 10, or 15...

6/3,K/24 (Item 2 from file: 370)

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Immunostimulatory DNA Sequences Necessary for Effective Intradermal Gene Immunization
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Text: Intramuscular (B1) or intradermal (B2) *administration* of pDNA expression vectors causes intracellular synthesis of the encoded proteins and induction of long-lasting cellular and humoral immune responses. Recently, we reported that...

...expression vectors encoding (beta) -galactosidase ((beta) -Gal) and containing a bacterial ampicillin resistance gene (ampR) produced a strong antibody response to (beta) -Gal (B3) . However, *subsequent* experiments showed that mice injected intradermally with a similar expression vector containing the kanamycin resistance gene (kanR) instead of ampR generated a weak antibody response...

...by the production of a distinctive cytokine profile [interleukin-2 (IL-2), tumor necrosis factor- (beta) (TNF- (beta)), and, mainly, interferon- (gamma) (IFN- (gamma))] by *antigen*-stimulated CD4 T cells (B6) . The CD4 splenocytes from pACB-Z-immunized mice generated large amounts of IFN- (gamma) and small amounts of IL-4...

...IFN-a, IFN- (beta) , and IFN- (gamma) from mouse splenocytes and human peripheral lymphocytes and to enhance natural killer cell activity. These ISS include the *following* *CpG*-containing hexamers: 5 (prime) -GACGTC-3 (prime) , 5 (prime) -AG-CGCT-3 (prime) , and 5 (prime) -AACGTT-3 (prime) (B7) . Two repeats of 5 (prime...

...that activated adherent splenocytes and enhanced natural killer cell

activity in vitro (B9) . Recently, Krieg et al. studied the effects of single-stranded oligonucleotides with *CpG* motifs on murine B lymphocyte activation (B10) . They found that cytosine methylation or the elimination of the *CpG* from the oligonucleotide abolished the lymphocyte stimulatory effect. The activation capability was attributed to a series of *CpG* -containing motifs that generally *follow* the formula 5 (prime) -Pur Pur CG Pyr Pyr-3 (prime) . *CpG*-enriched oligonucleotides induced not only B cell proliferation, but also the secretion of IL-6 and IFN- (gamma) (B11 ...

...al. showed that the stimulation of IFN- (gamma) synthesis by bacterial DNA is mediated by IL-12 and TNF-a (B20) . Therefore, keratinocytes and dermal *antigen*-presenting cells (APCs) transfected with ISS-containing pDNA could produce IFN-a and IL-12, which would then induce a T.inf(H)1 immune...

...Our findings indicate that immunogenic pDNA may be divided conceptually into two distinct units: a transcription unit that directs *antigen* synthesis and an adjuvant unit in the plasmid backbone that elicits the production of type-1 IFN and IL-12 in the transfected skin keratinocytes and APCs. For this reason, manipulation of the transcription unit within the pDNA to yield higher levels of *antigen* expression does not necessarily produce a stronger immune response. Both the localization and the precise sequence of the ISS within the plasmid backbone are also... ?